

US EPA ARCHIVE DOCUMENT

VOLUME 5 OF 7 OF SUBMISSION

STUDY TITLE

DIFENOCONAZOLE (CGA-169374)
ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374
RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK
BY GAS CHROMATOGRAPHY

DATA REQUIREMENT

EPA GUIDELINE NUMBER 171-4

APPROVED BY

ROBERT K. WILLIAMS

DATE STUDY COMPLETED

SEPTEMBER 15, 1988

PERFORMING LABORATORY

Residue Chemistry Department
Ciba Plant Protection
Ciba-Geigy Corporation
Greensboro, NC 27419

LABORATORY PROJECT IDENTIFICATION

AG-544

VOLUME 1 OF 1 OF STUDY

PAGE 1 OF 50

Ciba Plant Protection
Ciba-Geigy Corporation
Post Office Box 18300
Greensboro, NC 27419

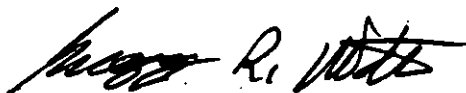
STATEMENT OF NO DATA CONFIDENTIALITY CLAIM

No claim of confidentiality is made for any information contained in this study on the basis of its falling within the scope of FIFRA Section 10 (d) (1) (A), (B) or (C).

Company: Ciba Plant Protection, Ciba-Geigy Corporation

Company Representative: Gregory R. Watson, Ph.D.

Title: Regulatory Manager



Signature

3-31-93

Date

These data are the property of Ciba Plant Protection, Ciba-Geigy Corporation and, as such, are considered confidential for all purposes other than compliance with FIFRA section 10.

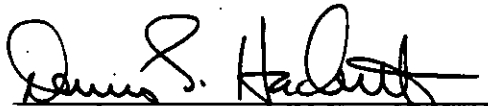
Submission of these data in compliance with FIFRA does not constitute a waiver of any right to confidentiality that may exist under any other statute or in any other country.

Submitter/Sponsor:
Ciba Plant Protection
Ciba-Geigy Corporation
Post Office Box 18300
Greensboro, NC 27419

STATEMENT CONCERNING GOOD LABORATORY PRACTICE

The analytical method contained within this volume was prepared in accordance with good and acceptable scientific practices.

Because Environmental Protection Agency's Good Laboratory Practice Standards (40 CFR 160, October 16, 1989) were not in effect at the completion date of this study, certification of compliance with GLPs is not applicable.



Dennis S. Hackett, Director
Biochemistry Department
Biochemistry Group
Agent of Submitter/Sponsor

6/4/93
Date

SUBMITTER/SPONSOR:
Ciba Plant Protection
Ciba-Geigy Corporation
Post Office Box 18300
Greensboro, NC 27419

TABLE OF CONTENTS

<u>TITLE</u>	<u>PAGE NO.</u>
AG-544: Analytical Method for the Determination of CGA-169374 in Dairy and Poultry Tissue, Eggs and Milk by Gas Chromatography	5
Appendix I: Residue Test Reports	40

Ciba Plant Protection
Ciba-Geigy Corporation
Post Office Box 18300
Greensboro, NC 27419

**ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374
RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK
BY GAS CHROMATOGRAPHY**

**ANALYTICAL METHOD NO. AG-544
BIOCHEMISTRY DEPARTMENT
AGRICULTURAL DIVISION
CIBA-GEIGY CORPORATION
GREENSBORO, NC 27419**

TABLE OF CONTENTS

	<u>Page No.</u>
I. INTRODUCTION/SUMMARY	
A. Scope.	5
B. Principle.	5
II. MATERIALS AND METHODS	
A. Apparatus.	5
B. Reagents	6
C. Analytical Procedure	6
1. Sample Preparation	6
2. Extraction	7
3. Partition Cleanup.	7
4. Silica SepPak Cleanup.	8
D. Instrumentation.	9
E. Interferences and Potential Problems	10
F. Confirmatory Techniques.	10
G. Time Required for Analysis	10
H. Modifications	10
I. Preparation of Standard CGA-169374 Solutions	11
J. Determination of Sample Residues	11
III. RESULTS AND DISCUSSION	13
IV. CONCLUSION	14
V. CERTIFICATION.	14
VI. TABLES AND FIGURES	
Table I. Gas Chromatographic Conditions for Analysis of CGA-169374	15

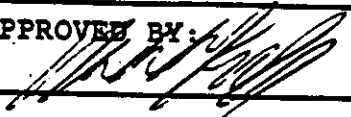
TABLE OF CONTENTS (Continued)

	<u>Page No.</u>
Table II. Typical Standardization Data for a CGA-169374 Standard Curve	16
Table III. Summary of Recovery Data for Various Dairy and Poultry Tissues, Eggs and Milk Fortified with CGA-169374	17
Table IV. Analysis of Goat Liver Samples Treated With ϕ - 14 C-CGA-169374	18
Figure 1. Chemical Structure of CGA-169374	19
Figure 2. Flow Diagram for Analytical Method AG-544	20
Figure 3. Typical Standard Chromatograms for CGA-169374	21
Figure 4. Typical Chromatograms for the Determination of CGA-169374 in Whole Milk	22
Figure 5. Typical Chromatograms for the Determination of CGA-169374 in Dairy Round	23
Figure 6. Typical Chromatograms for the Determination of CGA-169374 in Dairy Loin	24
Figure 7. Typical Chromatograms for the Determination of CGA-169374 in Dairy Perirenal Fat	25
Figure 8. Typical Chromatograms for the Determination of CGA-169374 in Dairy Omental Fat	25
Figure 9. Typical Chromatograms for the Determination of CGA-169374 in Dairy Kidney.	27

TABLE OF CONTENTS (Continued)

	<u>Page No.</u>
Figure 10. Typical Chromatograms for the Determination of CGA-169374 in Dairy Liver	28
Figure 11. Typical Chromatograms for the Determination of CGA-169374 in Dairy Blood	29
Figure 12. Typical Chromatograms for the Determination of CGA-169374 in Poultry Lean Meat	30
Figure 13. Typical Chromatograms for the Determination of CGA-169374 in Poultry Liver	31
Figure 14. Typical Chromatograms for the Determination of CGA-169374 in Poultry Fat	32
Figure 15. Typical Chromatograms for the Determination of CGA-169374 in Poultry Skin.	33
Figure 16. Typical Chromatograms for the Determination of CGA-169374 in Eggs	34
Figure 17. Chromatograms from the Analysis of Goat Liver Treated with ϕ - 14 C-CGA-169374	35

BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 5 of 35	METHOD NO. AG-544	SUBJECT ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
EDITION 9/15/88		
SUBMITTED BY: M. K. Ward		
		APPROVED BY: 

I. INTRODUCTION/SUMMARY

A. Scope

This method is used for the determination of parent residues of CGA-169374 (1-[[2-[2-chloro-4-(4-chlorophenoxy)phenyl]-4-methyl-1,3-dioxolan-2-yl]methyl]-1H-1,2,4-triazole) in animal tissues, eggs and milk. The limit of sensitivity of the method is 0.20 ng of CGA-169374. The limit of determination for CGA-169374 residues is 0.05 ppm of CGA-169374 (0.01 ppm of CGA-169374 in milk) as demonstrated by fortification experiments. The chemical name and structure of CGA-169374 are presented in Figure 1.

B. Principle

A representative sample is extracted by homogenizing for one minute with a Polytron homogenizer with 95:5 (v/v) acetonitrile:conc. ammonium hydroxide. After filtering, an aliquot of the extract is diluted with water and saturated sodium chloride and partitioned with hexane. The hexane fraction is partitioned with acetonitrile and the acetonitrile fraction is cleaned up on a silica gel SepPak. The final extract is analyzed by packed column gas chromatography using alkali flame ionization detection.

A flow diagram for the method is presented in Figure 2.

II. MATERIALS AND METHODS

A. Apparatus

- 1.0 Bottle, Boston round, 8-oz.
- 2.0 Filter paper, Whatman 2V, 24-cm., or equivalent
- 3.0 Flask, Erlenmeyer 250-ml.
- 4.0 Flask, round bottom, 50-ml. and 250-ml.
- 5.0 Funnel, filter, 10-cm.

BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 6 of 35	METHOD NO. AG-544	SUBJECT ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
EDITION		
SUBMITTED BY: M. K. Ward		
		APPROVED BY:

- 6.0 Funnel, separatory, 250-ml.
- 7.0 Polytron homogenizer, Brinkmann Instruments, or equivalent.
- 8.0 Rotary evaporator, Buchi, or equivalent.
- 9.0 SepPak, silica gel (Waters Assoc.)
- 10.0 Syringe, 20-ml, Luer-Lok.

B. Reagents

- 1.0 Acetone, reagent grade (Fisher, A18SK-4)
- 2.0 Acetonitrile, HPLC grade (Fisher, A998-4)
- 3.0 Ammonium hydroxide, concentrated (28-30%), reagent grade (Fisher, A-6695)
- 4.0 Hexane, HPLC grade (Fisher, H302-4)
- 5.0 95:5 (v/v) Acetonitrile:conc. ammonium hydroxide
- 6.0 Sodium chloride, reagent grade (Fisher, S-271)
- 7.0 Sodium chloride, saturated solution in distilled water
- 8.0 Toluene, reagent (Fisher, T324-4)
- 9.0 85:15 (v/v) Toluene:acetone
- 10.0 Water, distilled
- 11.0 CGA-169374 Analytical Standard, CIBA-GEIGY Corporation, P. O. Box 18300, Greensboro, N. C., 27419

C. Analytical Procedure

1.0 Sample Preparation

- 1.1 Beef and poultry meat, organ and fat samples are prepared by taking thin slices of the tissue from various sections of a partially frozen sample. The slices are then chopped into small pieces and mixed thoroughly before subsampling. Blood, eggs and milk are homogenized with the Polytron for a few seconds before the sample is withdrawn. Chicken skin, plus adhering fat, is prepared by placing alternate layers of crushed dry ice

BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 7 of 35	METHOD NO. AG-544	SUBJECT
EDITION		ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
SUBMITTED BY: M. K. Ward		
		APPROVED BY:

skin in a shallow pan. The pan is covered with aluminum foil and allowed to stand for 15-30 minutes at which time the frozen chicken skin is removed, cut into smaller pieces and combined in a blender with slightly less than equal portion of dry ice. After blending for several minutes, the mixture is poured into a plastic bag, loosely sealed and placed in a freezer until all of the carbon dioxide sublimates.

2.0 Extraction

- 2.1 Weigh a 20-g subsample of tissue, eggs or blood (40 g of milk) into a 16-oz. square, wide-mouth jar and add 185 ml of 95:5 (v/v) acetonitrile: concentrated ammonium hydroxide (165 ml for milk and 200 ml for fat). Homogenize the mixture for one minute with a Polytron homogenizer at medium speed.
- 2.2 Filter the homogenate through Whatman 2V filter paper into an 8-oz. Boston round bottle.

3.0 Partition Cleanup

- 3.1 Place a 50-ml aliquot (25 ml for milk) of the filtered extract solution in a 250-ml separatory funnel and add 125 ml of distilled water and 5 ml of saturated sodium chloride.
- 3.2 Partition the aqueous acetonitrile with 50 ml of hexane by shaking the separatory funnel for 30 seconds. Allow the layers to separate, draw the lower layer into a 250-ml Erlenmeyer flask leaving any emulsion in the separatory funnel, then add 5 ml of saturated sodium chloride to the separatory funnel to break most of the emulsion remaining. Draw off the aqueous layer and any remaining emulsion and combine it with the aqueous layer in the 250-ml Erlenmeyer flask.

BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 8 of 35	METHOD NO. AG-544	SUBJECT ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
EDITION		
SUBMITTED BY: M. K. Ward		
		APPROVED BY:

- 3.3 Pour the hexane layer from the top of the separatory funnel carefully, so as not to transfer any water droplets, into a second 250-ml separatory funnel.
- 3.4 Transfer the aqueous fraction back into the first 250-ml separatory funnel and partition with hexane a second time as in Section II.C.3.2.
- 3.5 Combine the hexane fractions in the second 250-ml separatory funnel. Partition the hexane with two 50-ml portions of acetonitrile, combine the acetonitrile fractions in a 250-ml round bottom flask and evaporate the contents of the flask to dryness on a rotary evaporator at a bath temperature of 40°C.

4.0 Silica SepPak Cleanup

- 4.1 Dissolve the residue in the round bottom flask in Section II.C.3.5 in 5 ml of toluene.
- 4.2 Connect a silica SepPak to the Luer fitting on a 20-ml Luer Lok syringe barrel and pre-wash the SepPak with 5 ml of toluene. Load the toluene solution from Section II.C.4.1 onto the SepPak. Discard the eluant.
- 4.3 Rinse the 250-ml round bottom flask with 5 ml of toluene and load the toluene onto the silica SepPak. Discard the eluant.
- 4.4 Elute the SepPak with 15 ml of 85:15 toluene: acetone and collect the eluant in a 50-ml round bottom flask.

BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 9 of 35	METHOD NO. AG-544	SUBJECT
EDITION		ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
SUBMITTED BY: M. K. Ward		
		APPROVED BY:

4.5 Evaporate the contents of the flask to dryness on a rotary evaporator at a bath temperature of 40°C. Dissolve the residue in the flask in 1.0 ml of toluene.

D. Instrumentation

1.0 Description and Operating Conditions

See Table I.

2.0 Calibration and Standardization

2.1 The GC system is calibrated with each analytical run by checking the retention time and detector response relative to previous runs. Retention times should not vary by more than 5% and detector response should not vary more than 10% between runs.

2.2 The GC system is standardized by injecting 5- μ l aliquots of standard solutions of CGA-169374 in a working range of 0.20-10.0 ng/injection. A linear regression function is generated from the data comparing detector response and ng injected. Alternately, a standard curve manually generated from plotting detector response versus ng injected may be used. Typical standard calculations are presented in Table II and typical standard chromatograms are shown in Figure 3.

2.3 As with any packed column GC system, the column should be sufficiently primed to give an optimal peak shape by deactivating any active sites on the column. This is accomplished by making several injections of sample matrix extracts until a constant peak shape and sensitivity are obtained for CGA-169374 prior to analyzing samples.

BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 10 of 35	METHOD NO. AG-544	SUBJECT ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
EDITION		
SUBMITTED BY: M. K. Ward		
		APPROVED BY:

E. Interferences and Potential Problems

- 1.0 Some interferences have been observed as a result of carry-over when large standard injections precede control and reagent blank injections. These interferences are particularly pronounced when "off-column" injection techniques are used. These problems can be minimized by properly maintaining the GC system (frequently changing the septum and glass wool at the head of the column), using direct "on-column" injection, and injecting samples and standards in a sequence where samples and standards of like concentration are adjacent.
- 2.0 Severe emulsions produced by some substrates in Section II.C.3.2 can be reduced or eliminated by addition of 5-10 ml portions of saturated sodium chloride. All samples of the set should be treated equally, however.
- 3.0 Analysis of control samples has shown no significant interferences at a screening level of 0.05 ppm in any substrate. No interferences have been observed in reagent blanks.

F. Confirmatory Techniques

None.

G. Time Required for Analysis

A skilled analyst can carry out the extraction, cleanup and analysis of a set of 4-6 samples in a 10-hour period including GC analysis.

H. Modifications

None.

BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 11 of 35	METHOD NO. AG-544	SUBJECT
EDITION		ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
SUBMITTED BY: M. K. Ward		APPROVED BY:

I. Preparation of Standard CGA-169374 Solutions

- 1.0 Weigh 100 mg of CGA-169374 analytical standard into a 100-ml volumetric flask and dilute to the mark with acetone.
- 2.0 Make serial dilutions of the 1 mg/ml standard solution in toluene to give a series of injection standards in a range of 0.04 to 2.0 ng/μl of CGA-169374.

J. Determination of Sample Residues

- 1.0 Inject 5-μl aliquots of sample extracts from Section II.C.4.5 into the gas chromatograph under the same conditions as for standards. Make appropriate dilutions of the samples in toluene (if necessary) to bring the sample peak heights within the range of the standard curve. Compare the peak heights of the unknown samples to the standard curve or enter the peak height into the least squares program to determine the nanograms of CGA-169374 in the injected aliquot. Typical chromatograms for control and procedural recovery samples are shown in Figures 4 through 16.

- 2.0 Calculate the residue results in terms of ppm of CGA-169374 by either equation (1) or (3) below:

$$(1) \text{ PPM CGA-169374} = \frac{\text{CGA-169374 found (ng)}}{\text{mg sample injected}} \times \frac{100}{R}$$

where mg sample injected is calculated as follows:

$$(2) \frac{\text{milligrams of sample extracted}}{\text{extraction volume including volume increase due to sample moisture (ml)}} \times \text{aliquot volume (ml)}$$

BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 12 of 35	METHOD NO. AG-544	SUBJECT
EDITION		ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
SUBMITTED BY: M. K. Ward		
		APPROVED BY:

$$X \frac{\text{injection volume } (\mu\text{l})}{\text{total volume of final injection solution } (\mu\text{l})}$$

Alternately, the residue results may be calculated by the following equation:

$$(3) \text{ PPM CGA-169374} = \frac{\text{CGA-169374 Found (ng)}}{\text{apparent mg injected}}$$

$$X \frac{V + (W \times M / 100)}{V} \times \frac{100}{R}$$

where apparent mg injected is determined by equation (2) above without including the volume increase due to sample moisture, V is the volume of the extraction solvent, W = weight of the crop sample extracted, M = percent moisture in the crop and 100 = conversion factor. R = percent recovery based on fortified controls taken through the procedure (See Section II.J.3).

This equation takes into account the increased volume of the extraction solvent due to the moisture contained in the sample, and the procedural recovery.

3.0 Fortification Experiments

The method is validated for each set of samples analyzed by including an untreated control sample and one or more control samples fortified prior to extraction with 0.05 ppm or more of CGA-169374 (0.01 ppm or more of CGA-169374 for milk).

BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 13 of 35	METHOD NO. AG-544	SUBJECT
EDITION		ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
SUBMITTED BY: M. K. Ward		
		APPROVED BY:

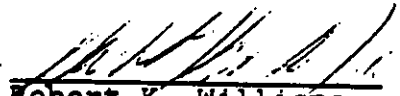
- 3.1 Add 1.0 ml of a 1.0-µg/ml standard solution of CGA-169374 to 20 g of sample, for a 0.05 ppm fortification, prior to addition of extraction solvent in Section II.C.2.1. Allow the sample to sit for a few minutes before adding the extraction solvent.
- 3.2 Analyze the samples through the procedures of the method as for treated samples.
- 3.3 Calculate the ppm of CGA-169374 in the samples excluding the 100/R recovery factor. Determine the recovery factor by first subtracting the detector response for CGA-169374, if any, in the control sample from the CGA-169374 response in the recovery sample. Then calculate the recovery factor expressed as a percentage (R%) by the equation:

$$(4) \quad R\% = \frac{\text{ppm CGA-169374 found}}{\text{ppm CGA-169374 added}} \times 100\%$$

III. RESULTS AND DISCUSSION

To date this method has been used for the analysis of control and CGA-169374-fortified control samples of dairy and poultry tissue, eggs and milk. Fortification levels ranged from 0.01 to 0.50 ppm. Recoveries averaged 99% with a standard deviation of 12% (N=52) and a range of 74-134%. At a screening level of 0.05 ppm (0.01 ppm for milk), no residues were found in any of the control samples. Results of these analyses are shown in Table III and are reported in AG-A 11060-01. The method has also been applied to control and ϕ - ^{14}C -CGA-169374 treated goat liver. The results of the goat liver analyses are shown in Table IV and are reported in AG-A 11077-01.

BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 14 of 35	METHOD NO. AG-544	SUBJECT ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
EDITION		
SUBMITTED BY: M. K. Ward		
		APPROVED BY:
<p>IV. CONCLUSION</p> <p>Analytical Method AG-544 is a valid and accurate method for the determination of parent residues of CGA-169374 in dairy and poultry tissue, eggs and milk.</p> <p>V. CERTIFICATION</p> <p>The reports and experimental results included in this study, Laboratory Project I. D. AG-544, are certified to be authentic accounts of the experiments.</p> <p><u>9/15/48</u> Date</p> <p> Robert K. Williams Research Scientist New Product Chemistry Biochemistry Dept. 919-292-7100, Ext. 2295</p> <p>AGRICULTURAL DIVISION CIBA-GEIGY CORPORATION POST OFFICE BOX 18300 GREENSBORO, NC 27419</p>		

BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 15 of 35	METHOD NO. AG-544	SUBJECT
EDITION		ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
SUBMITTED BY: M. K. Ward		
		APPROVED BY:

VI. TABLES AND FIGURES

TABLE I. GAS CHROMATOGRAPHIC CONDITIONS FOR ANALYSIS OF CGA-169374

Instrument: Hewlett-Packard 5880A Gas Chromatograph
with a 7673A Autosampler and N/P Detector

Carrier Gas: Helium, Flow Rate 30 ml/min.

Detector Gases: Hydrogen, Flow Rate 3 ml/min.
Air, Flow Rate 120 ml/min.

Column: 2 ft. x 2 mm, 3% OV-17 on
80-100 mesh GasChrom Q

Injection: 5- μ l, On-Column

Injector Temperature: 260°C

Detector Temperature: 280°C

Column Oven Temperature: 245°C

Retention Time: ~4.2 min.

Attenuation: 2 \uparrow 2

Chart Speed: 0.5 cm/min.

BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 16 of 35	METHOD NO. AG-544	SUBJECT ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
EDITION		
SUBMITTED BY: M. K. Ward		
		APPROVED BY:

TABLE II: TYPICAL STANDARDIZATION DATA FOR A CGA-169374 STANDARD CURVE

<u>Ng Injected</u>	<u>Peak Height (mm)</u>
0.20	2.5
0.50	6.0
1.0	12.
5.0	62.
7.5	94.
10.	124.

Linear Regression Analysis (TI-55 Calculator)

Slope: 12.46252 mm/ng
Intercept: -0.1821625 mm
Corr. Coeff.: 0.9999682

BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 17 of 35	METHOD NO. AG-544	SUBJECT ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
EDITION		
SUBMITTED BY: M. K. Ward		
		APPROVED BY:

TABLE III: SUMMARY OF RECOVERY DATA FOR VARIOUS DAIRY AND POULTRY
TISSUES, EGGS AND MILK FORTIFIED WITH CGA-169374
(AG-A 11060-01)

Substrate	Percent Recovery at Fortification (ppm) of:			
	0.01	0.05	0.10	0.5
<u>Dairy</u>				
Milk	134,102	118	--	111
Round	-	110,110	99	96
Loin	-	108,108	104	99
Perirenal Fat	-	95,108	97	95
Omental Fat	-	81,91	98	94
Kidney	-	89,89	96	102
Liver	-	121,121	109	109
Blood	-	110,98	99	93
<u>Poultry</u>				
Lean Meat (Breast and Thigh)	-	76,91	91	86
Liver	-	97,97	92	91
Fat	-	103,103	98	110
Skin	-	92,98	95	100
Eggs	-	74,81	81	83

Average recovery for milk at the 0.01 ppm screening level = 118%
(n = 2, S. D. = 16%).

Average recovery for all other substrates at the 0.05 ppm screening
level = 98% (n = 24, S. D. = 13%).

Average recovery for all substrates at all levels = 99% (n = 52,
S. D. = 12%).

BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 18 of 35	METHOD NO. AG-544	SUBJECT
EDITION		ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
SUBMITTED BY: M. K. Ward		
		APPROVED BY:

TABLE IV: ANALYSIS OF GOAT LIVER SAMPLES TREATED WITH
 ϕ - ^{14}C -CGA-169374¹ (AG-A 11077-01)

MGA No.: 145-01
 Test No.: M6-420-6,A
 Sample No.: G-6060052
 Total ppm²: 0.259

% of Total
 ^{14}C in Initial Extract³: 81%, 82%, 84%

% of Total
 ^{14}C in AG-544
 Final Fraction³: 16%, 16%, 17%

ppm of Total
 ^{14}C Residue
 Determined as
 Parent Residue
 of CGA-169374 by
 AG-544: <0.05, <0.05 <0.05

¹One lactating goat at the CIBA-GEIGY Research Station in Vero Beach, Florida was treated daily for ten consecutive days (June 17-June 27, 1986) with ϕ - ^{14}C -CGA-169374 at an average level of 4.17 ppm. See ABR-88087 for details. Analytical results are reported in AG-A 11077-01.

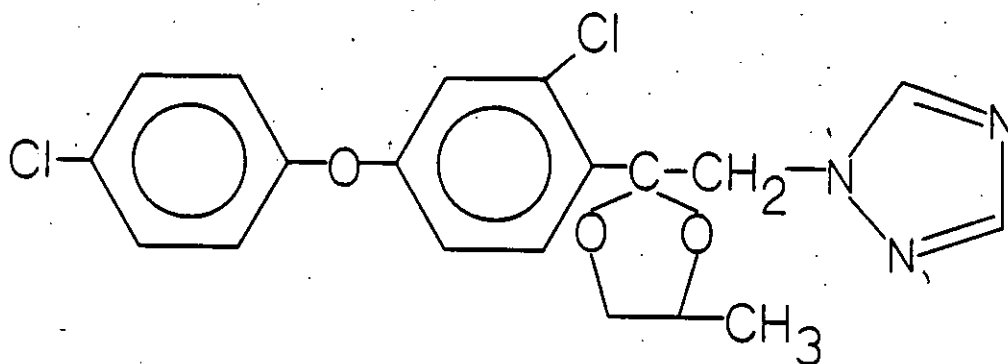
²Total ^{14}C in the goat liver was determined by combustion analysis and liquid scintillation counting of $^{14}\text{CO}_2$ liberated (SOP No. 4.67). See ABR-88087 for details.

³The extractability and percent of ^{14}C in the AG-544 final fraction were determined by liquid scintillation counting of aliquots of each solution (SOP No. 4.6).

BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 19 of 35	METHOD NO. AG-544	SUBJECT ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
EDITION		
SUBMITTED BY: M. K. Ward		
		APPROVED BY:

FIGURE 1. CHEMICAL STRUCTURE OF CGA-169374



CGA-169374

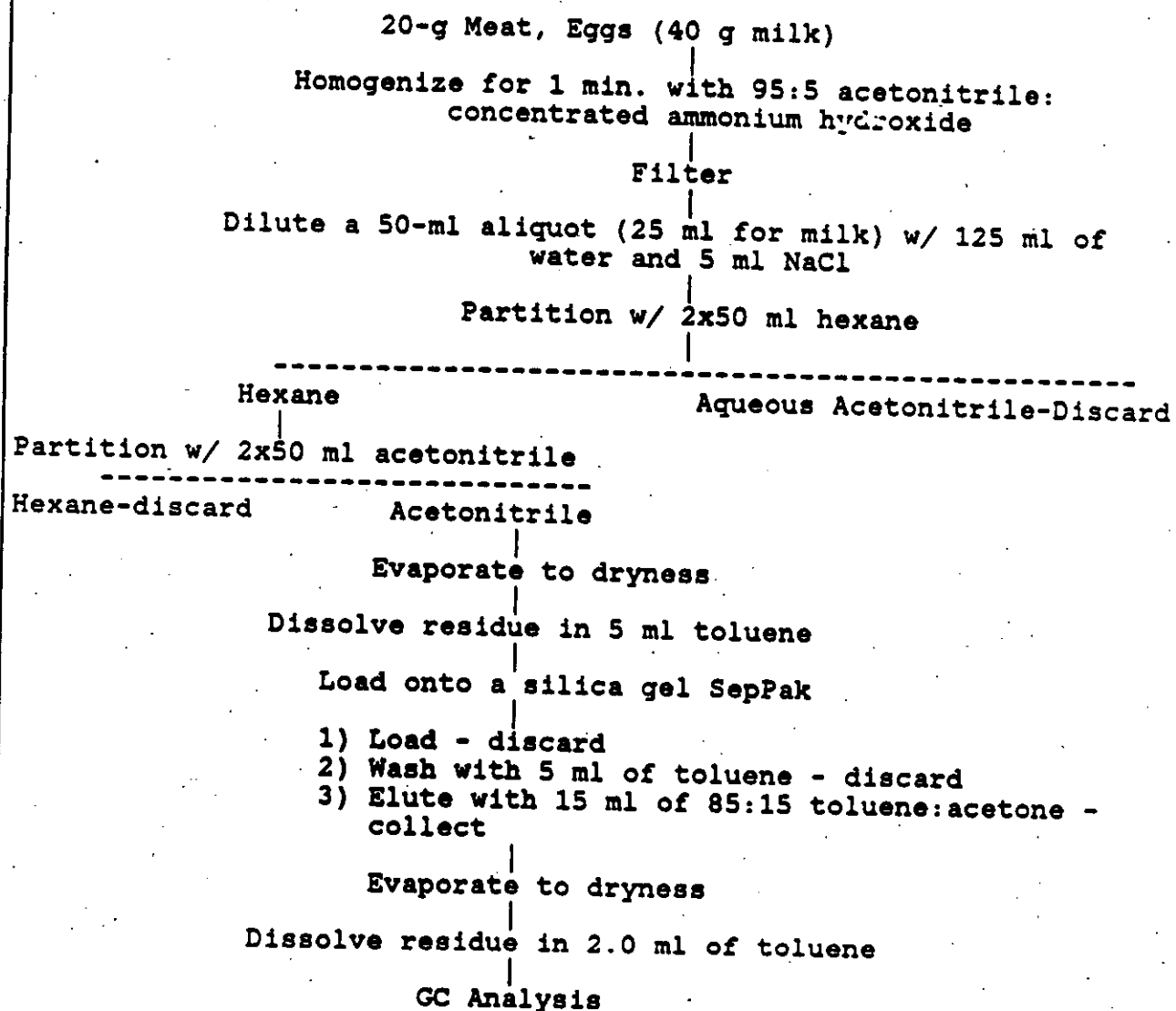
$C_{19}H_{17}N_3Cl_2O_3$ MW=406.27

1-[[2-[2-Chloro-4-(4-chlorophenoxy)phenyl]-4-methyl-1,3-dioxolan-2-yl]methyl]-1H-1,2,4-triazole

BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 20 of 35	METHOD NO. AG-544	SUBJECT ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
EDITION		
SUBMITTED BY: M. K. Ward		
		APPROVED BY:

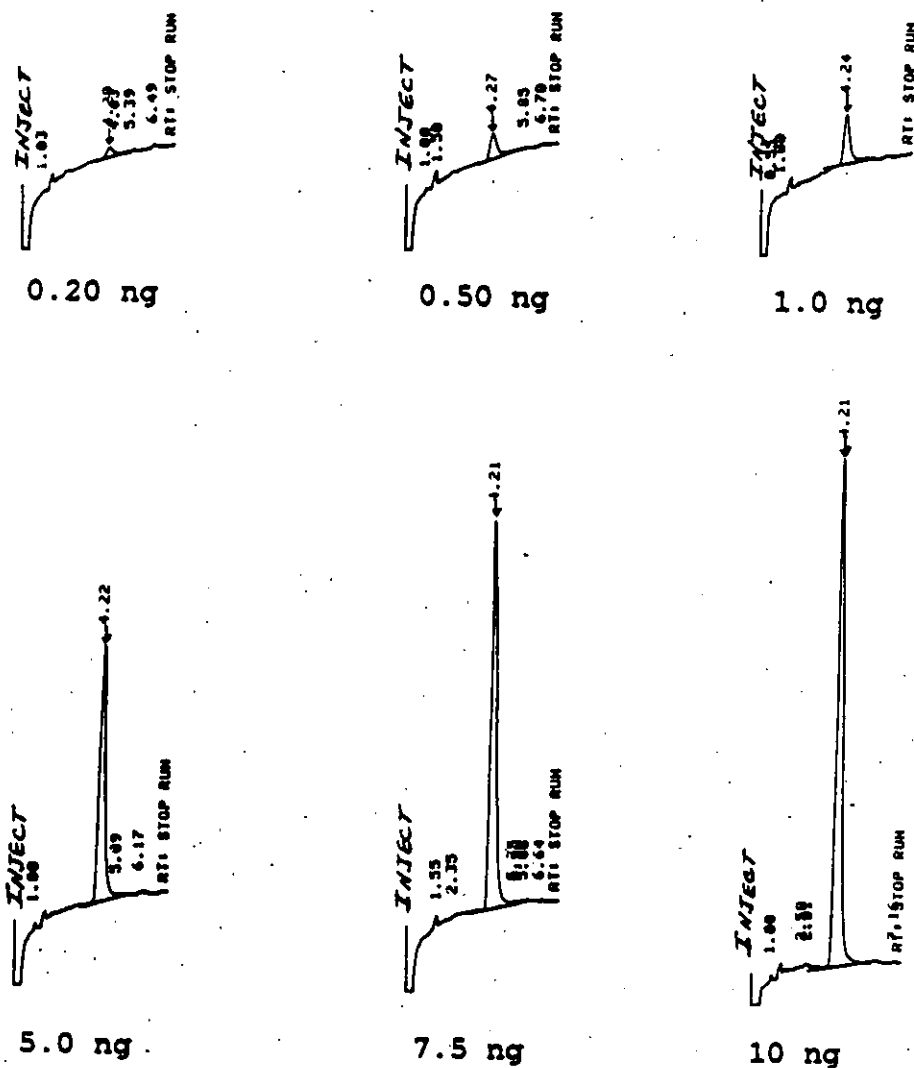
FIGURE 2. FLOW DIAGRAM FOR ANALYTICAL METHOD AG-544



BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 21 of 35	METHOD NO. AG-544	SUBJECT ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
EDITION		
SUBMITTED BY: M. K. Ward		
		APPROVED BY:

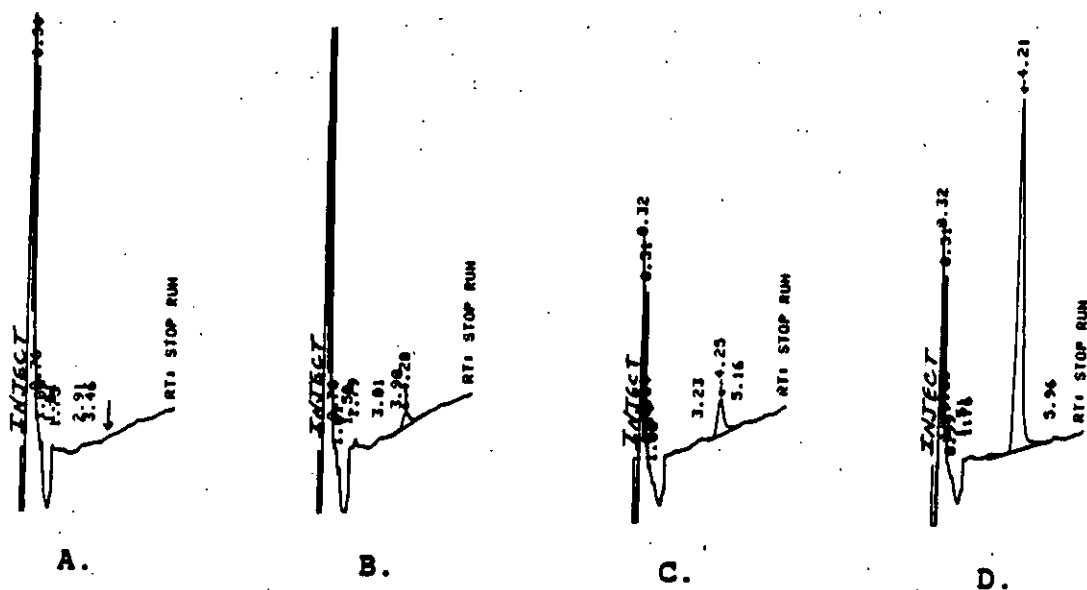
FIGURE 3. TYPICAL STANDARD CHROMATOGRAMS FOR CGA-169374



BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 22 of 35	METHOD NO. AG-544	SUBJECT ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
EDITION		
SUBMITTED BY: M. K. Ward		
		APPROVED BY:

FIGURE 4. TYPICAL CHROMATOGRAMS FOR THE DETERMINATION OF
CGA-169374 IN WHOLE MILK (AG-A 11060-01)

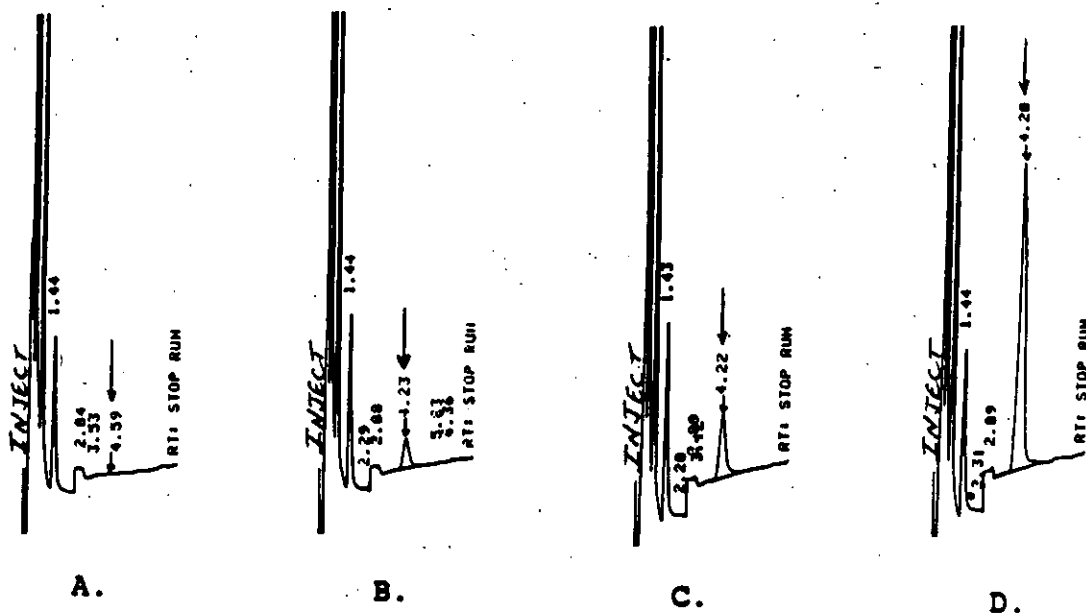


- A. Control whole milk, 25 mg injected, <0.20 ng found, <0.01 ppm.
- B. Control + 0.01 ppm CGA-169374, 25 mg injected, 0.26 ng found, 0.0102 ppm, 102% recovery.
- C. Control + 0.05 ppm CGA-169374, 12.5 mg injected, 0.74 ng found, 0.059 ppm, 118% recovery.
- D. Control + 0.50 ppm CGA-169374, 12.5 mg injected, 6.9 ng found, 0.55 ppm, 111% recovery.

BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 23 of 35	METHOD NO. AG-544	SUBJECT ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
EDITION		
SUBMITTED BY: M. K. Ward		
		APPROVED BY:

FIGURE 5. TYPICAL CHROMATOGRAMS FOR THE DETERMINATION OF CGA-169374 IN DAIRY ROUND (AG-A 11060-01)

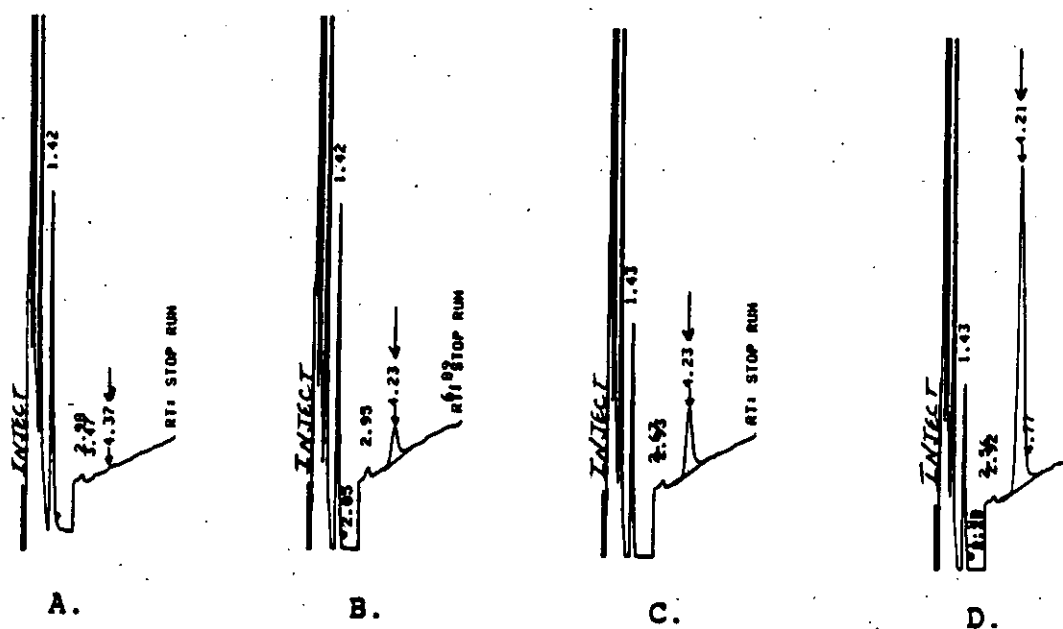


- A. Control dairy round, 12.5 mg injected, <0.20 ng found, <0.05 ppm.
- B. Control + 0.05 ppm CGA-169374, 12.5 mg injected, 0.69 ng found, 0.055 ppm, 110% recovery.
- C. Control + 0.10 ppm CGA-169374, 12.5 mg injected, 1.24 ng found, 0.099 ppm, 99% recovery.
- D. Control + 0.50 ppm CGA-169374, 12.5 mg injected, 6.0 ng found, 0.48 ppm, 96% recovery.

BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 24 of 35	METHOD NO. AG-544	SUBJECT ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
EDITION		
SUBMITTED BY: M. K. Ward		
		APPROVED BY:

FIGURE 6. TYPICAL CHROMATOGRAMS FOR THE DETERMINATION OF
CGA-169374 IN DAIRY LOIN (AG-A 11060-01)

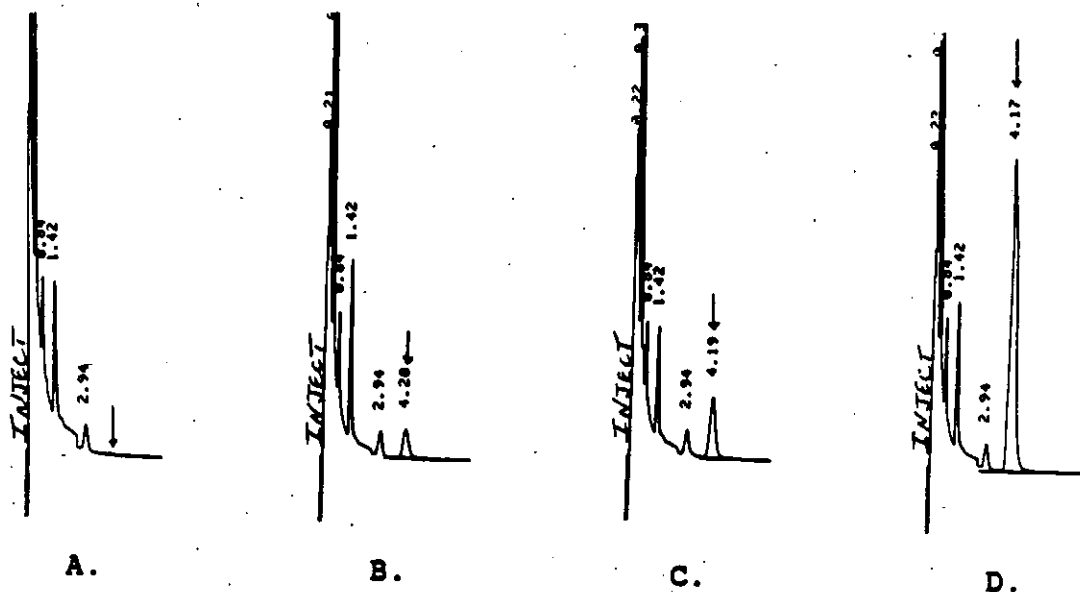


- Control dairy loin, 12.5 mg injected, <0.20 ng found, <0.05 ppm.
- Control + 0.05 ppm CGA-169374, 12.5 mg injected, 0.67 ng found, 0.054 ppm, 108% recovery.
- Control + 0.10 ppm CGA-169374, 12.5 mg injected, 1.30 ng found, 0.104 ppm, 104% recovery.
- Control + 0.50 ppm CGA-169374, 12.5 mg injected, 6.2 ng found, 0.50 ppm, 99% recovery.

BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 25 of 35	METHOD NO. AG-544	SUBJECT ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
EDITION		
SUBMITTED BY: M. K. Ward		
		APPROVED BY:

FIGURE 7. TYPICAL CHROMATOGRAMS FOR THE DETERMINATION OF CGA-169374 IN DAIRY PERIRENAL FAT (AG-A 11060-01)

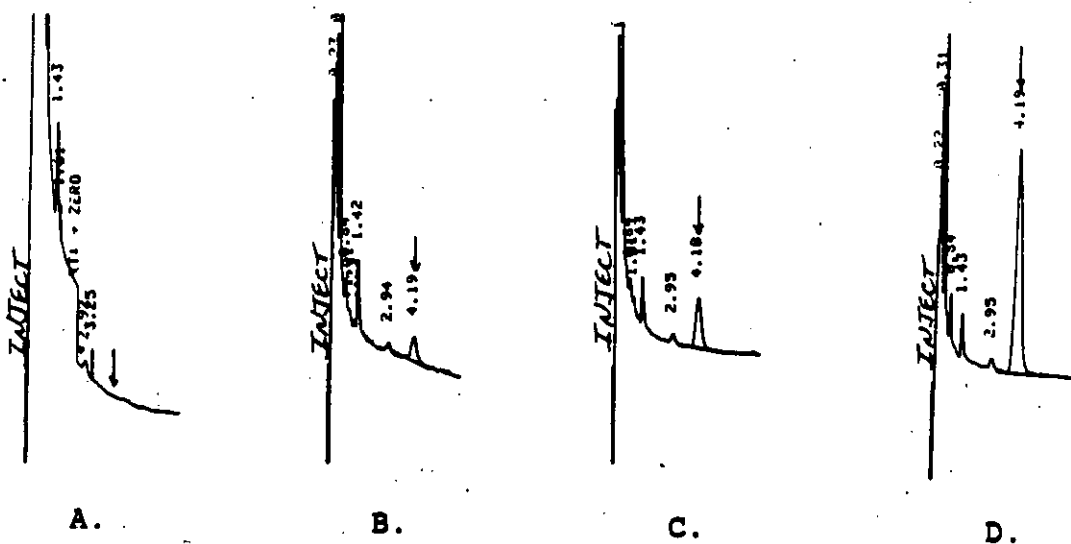


- A. Control dairy perirenal fat, 12.5 mg injected, <0.20 ng found, <0.05 ppm.
- B. Control + 0.05 ppm CGA-169374, 12.5 mg injected, 0.60 ng found, 0.048 ppm, 95% recovery.
- C. Control + 0.10 ppm CGA-169374, 12.5 mg injected, 1.21 ng found, 0.097 ppm, 97% recovery.
- D. Control + 0.50 ppm CGA-169374, 12.5 mg injected, 5.9 ng found, 0.48 ppm, 95% recovery.

BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 26 of 35	METHOD NO. AG-544	SUBJECT ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
EDITION		
SUBMITTED BY: M. K. Ward		
		APPROVED BY:

FIGURE 8. TYPICAL CHROMATOGRAMS FOR THE DETERMINATION OF CGA-169374 IN DAIRY OMENTAL FAT (AG-A 11060-01)

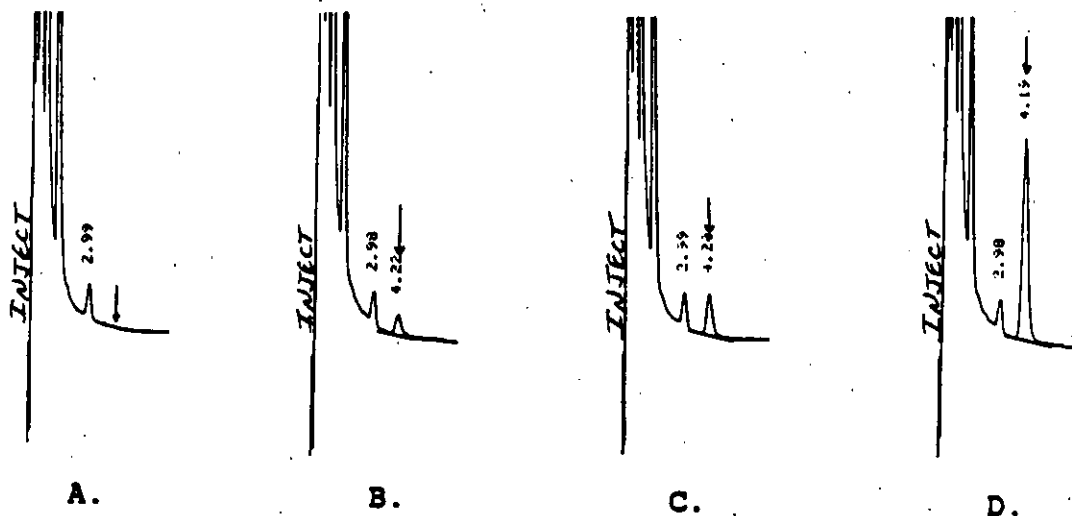


- A. Control dairy omental fat, 12.5 mg injected, <0.20 ng found, <0.05 ppm.
- B. Control + 0.05 ppm CGA-169374, 12.5 mg injected, 0.57 ng found, 0.045 ppm, 91% recovery.
- C. Control + 0.10 ppm CGA-169374, 12.5 mg injected, 1.22 ng found, 0.098 ppm, 98% recovery.
- D. Control + 0.50 ppm CGA-169374, 12.5 mg injected, 5.9 ng found, 0.47 ppm, 94% recovery.

BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 27 of 35	METHOD NO. AG-544	SUBJECT ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
EDITION		
SUBMITTED BY: M. K. Ward		
		APPROVED BY:

FIGURE 9. TYPICAL CHROMATOGRAMS FOR THE DETERMINATION OF
CGA-169374 IN DAIRY KIDNEY (AG-A 11060-01)

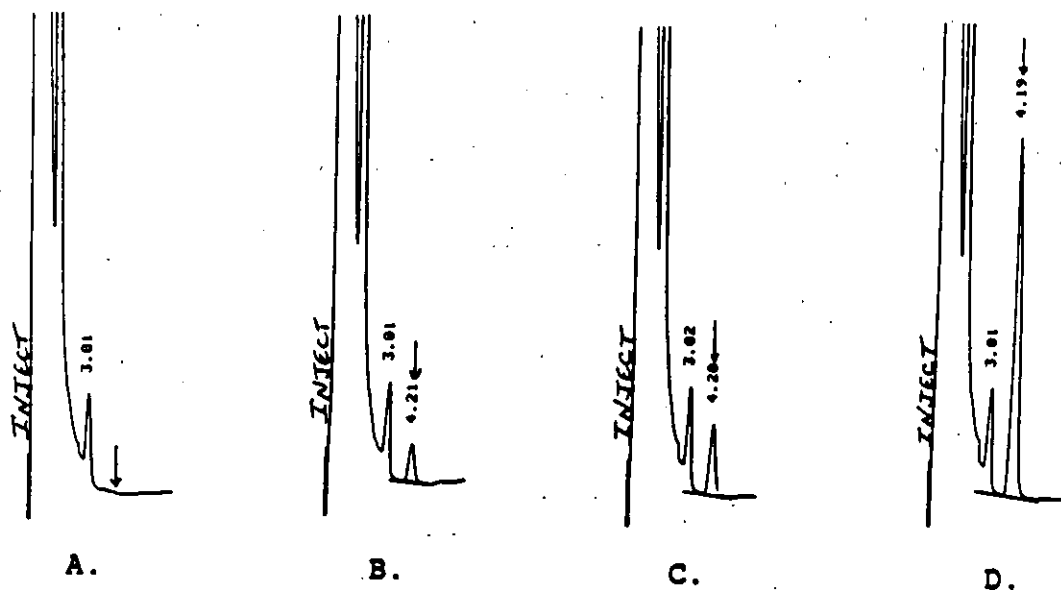


- A. Control dairy kidney, 12.5 mg injected, <0.20 ng found, <0.05 ppm.
- B. Control + 0.05 ppm CGA-169374, 12.5 mg injected, 0.55 ng found, 0.044 ppm, 89% recovery.
- C. Control + 0.10 ppm CGA-169374, 12.5 mg injected, 1.20 ng found, 0.096 ppm, 96% recovery.
- D. Control + 0.50 ppm CGA-169374, 12.5 mg injected, 6.4 ng found, 0.51 ppm, 102% recovery.

BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 28 of 35	METHOD NO. AG-544	SUBJECT ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
EDITION		
SUBMITTED BY: M. K. Ward		
		APPROVED BY:

FIGURE 10. TYPICAL CHROMATOGRAMS FOR THE DETERMINATION OF
CGA-169374 IN DAIRY LIVER (AG-A 11060-01)

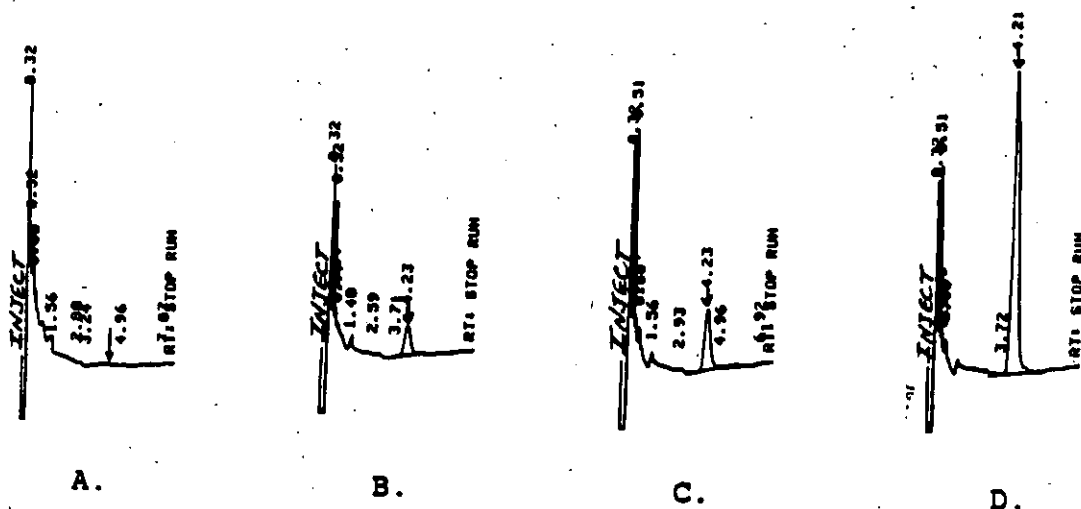


- A. Control dairy liver, 12.5 mg injected, <0.20 ng found, <0.05 ppm.
- B. Control + 0.05 ppm CGA-169374, 12.5 mg injected, 0.76 ng found, 0.061 ppm, 121% recovery.
- C. Control + 0.10 ppm CGA-169374, 12.5 mg injected, 1.36 ng found, 0.109 ppm, 109% recovery.
- D. Control + 0.50 ppm CGA-169374, 12.5 mg injected, 6.8 ng found, 0.54 ppm, 109% recovery.

BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 29 of 35	METHOD NO. AG-544	SUBJECT ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
EDITION		
SUBMITTED BY: M. K. Ward		
		APPROVED BY:

FIGURE 11. TYPICAL CHROMATOGRAMS FOR THE DETERMINATION OF
CGA-169374 IN DAIRY BLOOD (AG-A 11060-01)

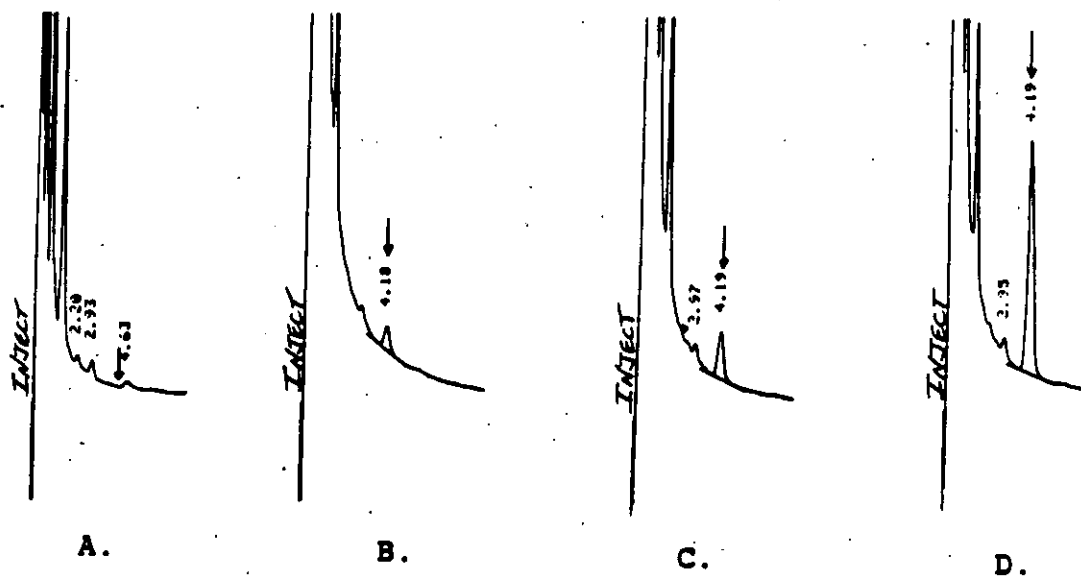


- A. Control blood, 12.5 mg injected, <0.20 ng found, <0.05 ppm.
- B. Control + 0.05 ppm CGA-169374, 12.5 mg injected, 0.61 ng found, 0.049 ppm, 98% recovery.
- C. Control + 0.10 ppm CGA-169374, 12.5 mg injected, 1.24 ng found, 0.099 ppm, 99% recovery.
- D. Control + 0.50 ppm CGA-169374, 12.5 mg injected, 5.8 ng found, 0.47 ppm, 93% recovery.

BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 30 of 35	METHOD NO. AG-544	SUBJECT ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
EDITION		
SUBMITTED BY: M. K. Ward		
		APPROVED BY:

FIGURE 12. TYPICAL CHROMATOGRAMS FOR THE DETERMINATION OF
CGA-169374 IN POULTRY LEAN MEAT (AG-A 11060-01)

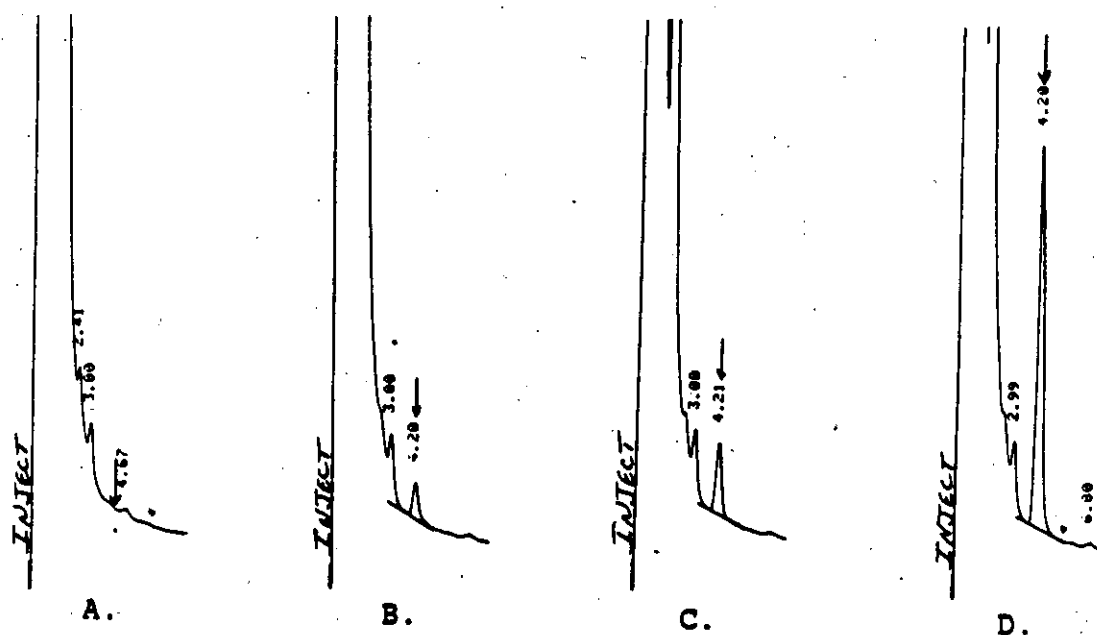


- A. Control chicken lean meat, 12.5 mg injected, <0.20 ng found, <0.05 ppm.
- B. Control + 0.05 ppm CGA-169374, 12.5 mg injected, 0.57 ng found, 0.046 ppm, 91% recovery.
- C. Control + 0.10 ppm CGA-169374, 12.5 mg injected, 1.13 ng found, 0.091 ppm, 91% recovery.
- D. Control + 0.50 ppm CGA-169374, 12.5 mg injected, 5.3 ng found, 0.43 ppm, 86% recovery.

BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 31 of 35	METHOD NO. AG-544	SUBJECT ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
EDITION		
SUBMITTED BY: M. K. Ward		
		APPROVED BY:

FIGURE 13. TYPICAL CHROMATOGRAMS FOR THE DETERMINATION OF CGA-169374 IN POULTRY LIVER (AG-A 11060-01)

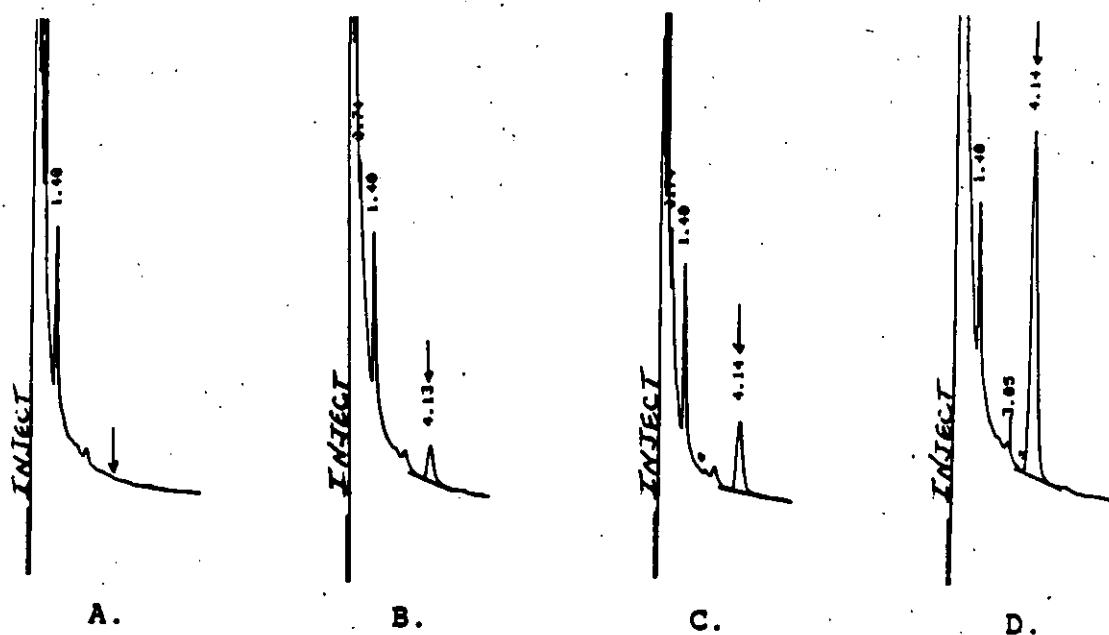


- A. Control chicken liver, 12.5 mg injected, <0.20 ng found, <0.05 ppm.
- B. Control + 0.05 ppm CGA-169374, 12.5 mg injected, 0.61 ng found, 0.049 ppm, 97% recovery.
- C. Control + 0.10 ppm CGA-169374, 12.5 mg injected, 1.15 ng found, 0.092 ppm, 92% recovery.
- D. Control + 0.50 ppm CGA-169374, 12.5 mg injected, 5.7 ng found, 0.46 ppm, 91% recovery.

BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 32 of 35	METHOD NO. AG-544	SUBJECT ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
EDITION		
SUBMITTED BY: M. K. Ward		
		APPROVED BY:

FIGURE 14. TYPICAL CHROMATOGRAMS FOR THE DETERMINATION OF
CGA-169374 IN POULTRY FAT (AG-A 11060-01)

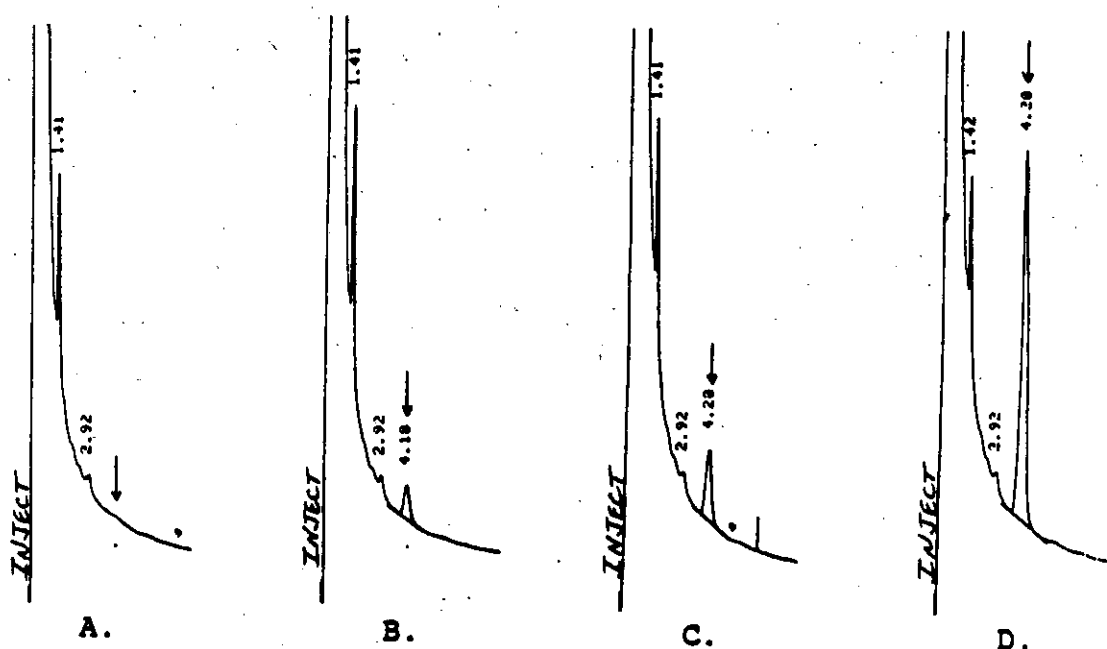


- A. Control chicken fat, 12.5 mg injected, <0.20 ng found, <0.05 ppm.
- B. Control + 0.05 ppm CGA-169374, 12.5 mg injected, 0.64 ng found, 0.051 ppm, 103% recovery.
- C. Control + 0.10 ppm CGA-169374, 12.5 mg injected, 1.22 ng found, 0.098 ppm, 98% recovery.
- D. Control + 0.50 ppm CGA-169374, 12.5 mg injected, 6.9 ng found, 0.55 ppm, 110% recovery.

BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 33 of 35	METHOD NO. AG-544	SUBJECT ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
EDITION		
SUBMITTED BY: M. K. Ward		
		APPROVED BY:

FIGURE 15. TYPICAL CHROMATOGRAMS FOR THE DETERMINATION OF
CGA-169374 IN POULTRY SKIN (AG-A 11060-01)

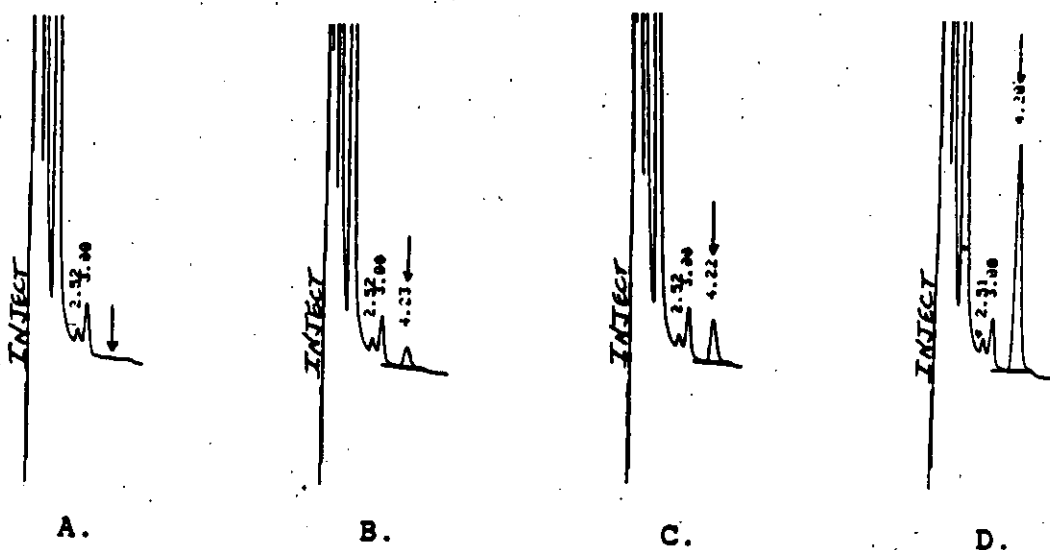


- A. Control chicken skin, 12.5 mg injected, <0.20 ng found, <0.05 ppm.
- B. Control + 0.05 ppm CGA-169374, 12.5 mg injected, 0.58 ng found, 0.046 ppm, 92% recovery.
- C. Control + 0.10 ppm CGA-169374, 12.5 mg injected, 1.19 ng found, 0.095 ppm, 95% recovery.
- D. Control + 0.50 ppm CGA-169374, 12.5 mg injected, 6.2 ng found, 0.50 ppm, 100% recovery.

BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 34 of 35	METHOD NO. AG-544	SUBJECT ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
EDITION		
SUBMITTED BY: M. K. Ward		
		APPROVED BY:

FIGURE 16. TYPICAL CHROMATOGRAMS FOR THE DETERMINATION OF
CGA-169374 IN EGGS (AG-A 11060-01)

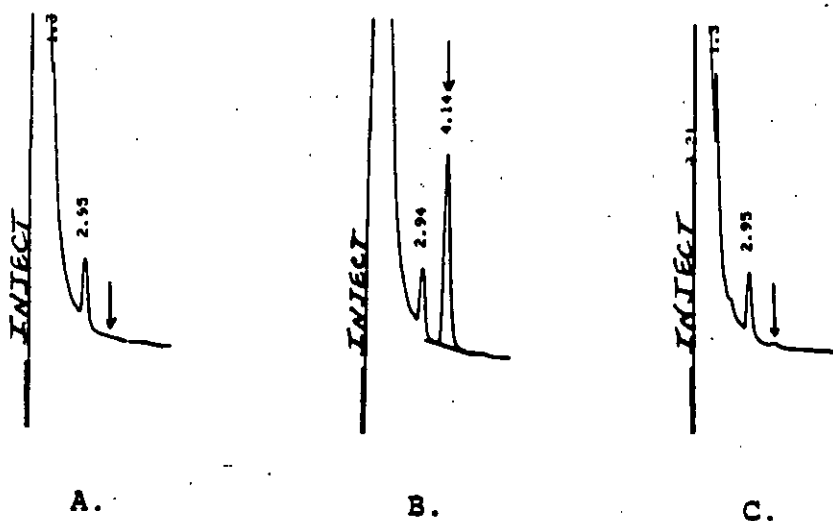


- A. Control whole eggs, 12.5 mg injected, <0.20 ng found, <0.05 ppm.
- B. Control + 0.05 ppm CGA-169374, 12.5 mg injected, 0.51 ng found, 0.041 ppm, 81% recovery.
- C. Control + 0.10 ppm CGA-169374, 12.5 mg injected, 1.01 ng found, 0.081 ppm, 81% recovery.
- D. Control + 0.50 ppm CGA-169374, 12.5 mg injected, 5.2 ng found, 0.41 ppm, 83% recovery.

BIOCHEMISTRY DEPARTMENT
CIBA-GEIGY CORPORATION
GREENSBORO, N.C.

PAGE 35 of 35	METHOD NO. AG-544	SUBJECT ANALYTICAL METHOD FOR THE DETERMINATION OF CGA-169374 RESIDUES IN DAIRY AND POULTRY TISSUE, EGGS AND MILK BY GAS CHROMATOGRAPHY
EDITION		
SUBMITTED BY: M. K. Ward		
		APPROVED BY:


FIGURE 17. CHROMATOGRAMS FROM THE ANALYSIS OF GOAT LIVER
TREATED WITH ϕ - 14 C-CGA-169374 (AG-A 11077-01)



- A. Control goat liver, 12.5 mg injected, <0.20 ng found, <0.05 ppm.
- B. Control goat liver + 0.30 ppm CGA-169374, 12.5 mg injected, 3.7 ng found, 0.30 ppm, 100% recovery.
- C. ϕ - 14 C-CGA-169374 treated goat liver, 12.5 mg injected, <0.20 ng found, <0.05 ppm

APPENDIX I
RESIDUE TEST REPORTS

Ciba Plant Protection
Ciba-Geigy Corporation
Post Office Box 18300
Greensboro, NC 27419

Compound(s) and Formulations(s): ϕ - ¹⁴ C-CGA-169374 97% Radiochemical Purity (GAN-IX-5)		Commodity: Goat	Substrate: Liver																							
C-G Rep.:	Plot Location:	Growth Stages Sampled:	Cooperator Name and Address:																							
Soil Type:	Date Planted:																									
Treatment Rates:		Method of Application:	Equipment:	Vol. per Acre:																						
Dates of Application:		Sampling Date(s):																								
Other Materials Applied:		Sample Care Before Storage:																								
Storage Information: Frozen	No. of Analyses: 5	Plot Maintenance, i.e., Cultivation, Irrigation, etc.:																								
<p>Summary of Results: At the CIBA-GEIGY Research Facility in Vero Beach, Florida, a lactating goat was treated with ten daily consecutive oral doses of ϕ-¹⁴C-CGA-169374 (48.6 μCi/mg) at a level equivalent to approximately 4 ppm in the feed. Twenty-two hours after the last dose, the goat was sacrificed and samples of the tissues were collected. Liver from this goat was analyzed for parent CGA-169374 residues by AG-544 and the results are shown below along with the percent of total radioactive residue present in the goat liver extract and the final fraction (used for GC analysis) as determined by liquid scintillation counting of aliquots from these solutions.</p> <table border="1"><thead><tr><th>Test No.</th><th>Sample</th><th>Total ppm ¹⁴C</th><th>Replicate</th><th>% ¹⁴C in Extract</th><th>% ¹⁴C (ppm) in Final Fraction</th><th>GC Results (ppm)</th></tr></thead><tbody><tr><td rowspan="3">M6-420-6A*</td><td rowspan="3">G6060052</td><td rowspan="3">0.259</td><td>1</td><td>81%</td><td>16% (0.041)</td><td><0.05</td></tr><tr><td>2</td><td>82%</td><td>16% (0.041)</td><td><0.05</td></tr><tr><td>3</td><td>84%</td><td>17% (0.043)</td><td><0.05</td></tr></tbody></table> <p>*See ABR-88087 for details.</p>					Test No.	Sample	Total ppm ¹⁴ C	Replicate	% ¹⁴ C in Extract	% ¹⁴ C (ppm) in Final Fraction	GC Results (ppm)	M6-420-6A*	G6060052	0.259	1	81%	16% (0.041)	<0.05	2	82%	16% (0.041)	<0.05	3	84%	17% (0.043)	<0.05
Test No.	Sample	Total ppm ¹⁴ C	Replicate	% ¹⁴ C in Extract	% ¹⁴ C (ppm) in Final Fraction	GC Results (ppm)																				
M6-420-6A*	G6060052	0.259	1	81%	16% (0.041)	<0.05																				
			2	82%	16% (0.041)	<0.05																				
			3	84%	17% (0.043)	<0.05																				
Date Received:	Date Extracted: 8/22/88	Date Analyzed: 8/23/88	Analyst: MKW																							
Method of Analysis: AG-544																										
Analysis Approved By: 			Date Approved: 9/12/88																							

ts/SEPT/AGA-11077/F

CIBA-GEIGY Corporation

Compound(s)			Goat Liver		Residue - ppm
ϕ - ¹⁴ C-CGA-169374			Substrate	Replicate	
Sample Code	ppm	Application Date(s)	Sample Date(s)	ϕ - ¹⁴ C-CGA-169374	
Goat #35* (Liver B)	0.00	-	6/4/85	-	
G060052 (Liver)	4.17**	6/18-27/86	6/27/86	1 2 3	
				<0.05 <0.05 <0.05 <0.05	

Comments: Residue results are not corrected for control values. Residue results are corrected for procedural recoveries of <100% and for moisture content of 80% by formula in Section 11.J.2 of AG-594.

*Control goat #35: Test No. M6-161-5A (See ABR-85076 for details).

**Average daily dose based on the feed.


ts/SEPT/AGA-11077/F

AG-A No. 11077-01

Page 3 of 3

Comments: Recovery samples are fortified prior to extraction.
*Liver ppm values are corrected for 80% moisture content by formula in Section II.J.2 of AG-544.

TS/SEPT/AGA-11077/F

Compound(s) and Formulations(s): CGA-169374, S85-0812, 94.5%		Commodity:		Substrate: Dairy and Poultry Tissue, Egg and Milk Control Samples	
C-G Rep.: --	Plot Location: --	Growth Stages Sampled: --		Cooperator Name and Address: --	
Soil Type: --	Date Planted: --				
Treatment Rate(s): --		Method of Application: --		Equipment: --	Vol. per Acre: --
Date(s) of Application: --		Sampling Date(s): --			
Other Materials Applied: --		Sample Care Before Storage: --			
Storage Information: Frozen	No. of Analyses: 65	Plot Maintenance, i.e., Cultivation, Irrigation, etc.: --			
Summary of Results: See page 2 for summary of recovery data.					
Date Received: --	Date Extracted: 5/88	Date Analyzed: 6/88 - 8/88		Analyst: MKW	
Method of Analysis: AG-544					
Analysis Approved By: 				Date Approved: 9/15/88	

nt/SEPTEMBER

Summary of Results: Method trials for Analytical Method AG-544 validation were conducted on dairy and poultry tissue, eggs and milk control and fortified control samples with the following results:

<u>AG-A</u>	<u>Substrate</u>	<u>Sample</u>	<u>Fortification Level (ppm)</u>	<u>Average Recovery \pm SD (%)</u>	
10242-02	Whole milk	1-31-B	0.01 - 0.50	116 \pm 12	(n = 4)
10243-02	Dairy blood	1-16-B	0.05 - 0.50	100 \pm 6	(n = 4)
10244-02	Dairy loin	1-21-B	0.05 - 0.50	105 \pm 4	(n = 4)
10244-02	Dairy round	1-20-B	0.05 - 0.50	104 \pm 6	(n = 4)
10244-02	Dairy liver	1-34-B	0.05 - 0.50	115 \pm 6	(n = 4)
10244-02	Dairy perirenal fat	1-18-B	0.05 - 0.50	99 \pm 5	(n = 4)
10244-02	Dairy omental fat	1-19-B	0.05 - 0.50	91 \pm 6	(n = 4)
10244-02	Dairy kidney	1-22-B	0.05 - 0.50	94 \pm 5	(n = 4)
8667-01	Chicken lean meat	1-23-B	0.05 - 0.50	86 \pm 6	(n = 4)
10106-01	Whole eggs	1-15-A	0.05 - 0.50	80 \pm 3	(n = 4)
8667-01	Chicken skin (composited)	1-20-A	0.05 - 0.50	96 \pm 3	(n = 4)
		1-24-A			
Commercial	Chicken liver	--	0.05 - 0.50	94 \pm 3	(n = 4)
8667-01	Chicken fat (composited)	1-17-A	0.05 - 0.50	104 \pm 4	(n = 4)
		1-21-A			
		1-25-A			

Average recovery for all substrates at all levels = 99 \pm 12% (n = 52).

RESIDUE RECOVERY REPORT

Page 3 of 7AG-A No. 11060-01

Compound(s)		Substrate(s)					
CGA-169374		Control Whole Milk, Dairy Blood and Loin					
		Pesticide Added					
		CGA-169374					
SUBSTRATE	SAMPLE WEIGHT mg	AMOUNT ADDED		FOUND			
		ng	ppm	TOTAL		NET	
				ng	ppm*	ppm	%
Control Whole Milk (1-31-B)	25	0.0	0.00	<0.20	<0.01	<0.01	
CGA-169374	25	0.25	0.01	0.34	0.0134	0.0134	134
CGA-169374	25	0.25	0.01	0.26	0.0102	0.0102	102
CGA-169374	12.5	1.25	0.10	0.74	0.059	0.059	118
CGA-169374	12.5	6.2	0.50	6.9	0.55	0.55	111
Control Blood (1-16-B)	12.5	0.0	0.00	<0.20	<0.05	<0.05	
CGA-169374	12.5	0.62	0.05	0.69	0.055	0.055	110
CGA-169374	12.5	0.62	0.05	0.61	0.049	0.049	98
CGA-169374	12.5	1.25	0.10	1.24	0.099	0.099	99
CGA-169374	12.5	6.2	0.50	5.8	0.47	0.47	93
Control Dairy Loin (1-21-B)	12.5	0.0	0.00	<0.20	<0.05	<0.05	
CGA-169374	12.5	0.62	0.05	0.67	0.054	0.054	108
CGA-169374	12.5	0.62	0.05	0.67	0.054	0.054	108
CGA-169374	12.5	1.25	0.10	1.30	0.104	0.104	104
CGA-169374	12.5	6.2	0.50	6.2	0.50	0.50	99

Comments: Recovery samples fortified prior to extraction.

*Milk ppm values are corrected for 87% moisture content, and blood and loin are corrected for 80% moisture content using formula in Section II.J.2 of AG-544.

mt/SEPTEMBER

RESIDUE RECOVERY REPORT

AG-A No. 11060-01

Page 4 of 7

Compound(s)		Substrate(s)					
CGA-169374		Control Dairy Round Meat, Liver and Perirenal Fat					
		Pesticide Added					
		CGA-169374					
SUBSTRATE	SAMPLE WEIGHT g	AMOUNT ADDED		FOUND			
		ng	ppm	TOTAL		NET	
				ng	ppm*	ppm	%
Control Dairy Round Meat (1-20-B)	12.5	0.0	0.00	<0.20	<0.05	<0.05	
CGA-169374	12.5	0.62	0.05	0.69	0.055	0.055	110
CGA-169374	12.5	0.62	0.05	0.69	0.055	0.055	110
CGA-169374	12.5	1.25	0.10	1.24	0.099	0.099	99
CGA-169374	12.5	6.2	0.50	6.0	0.48	0.48	96
Control Dairy Liver (1-34-B)	12.5	0.0	0.00	<0.20	<0.05	<0.05	
CGA-169374	12.5	0.62	0.05	0.76	0.061	0.061	121
CGA-169374	12.5	0.62	0.05	0.76	0.061	0.061	121
CGA-169374	12.5	1.25	0.10	1.36	0.109	0.109	109
CGA-169374	12.5	6.2	0.50	6.8	0.54	0.54	109
Control Dairy Perirenal Fat (1-18-B)	12.5	0.0	0.00	<0.20	<0.05	<0.05	
CGA-169374	12.5	0.62	0.05	0.60	0.048	0.048	95
CGA-169374	12.5	0.62	0.05	0.67	0.054	0.054	108
CGA-169374	12.5	1.25	0.10	1.21	0.097	0.097	97
CGA-169374	12.5	6.2	0.50	5.9	0.48	0.48	95

Comments: Recovery samples fortified prior to extraction.

*Dairy round meat and liver ppm values are corrected for 80% moisture content by formula in Section II.J.2 of AG-544.

mt/SEPTEMBER

RESIDUE RECOVERY REPORT

Page 5 of 7AG-A No. 11060-01

Compound(s)		Substrate(s) Control Dairy Omental Fat and Kidney and Chicken Lean Meat					
CGA-169374		Pesticide Added CGA-169374					
SUBSTRATE	SAMPLE WEIGHT	AMOUNT ADDED		FOUND			
		ng	ppm	TOTAL		NET	
				ng	ppm*	ppm	%
Control Dairy Omental Fat (1-19-B)	12.5	0.0	0.00	<0.20	<0.05	<0.05	
CGA-169374	12.5	0.62	0.05	0.51	0.041	0.041	81
CGA-169374	12.5	0.62	0.05	0.57	0.045	0.045	91
CGA-169374	12.5	1.25	0.10	1.22	0.098	0.098	98
CGA-169374	12.5	6.2	0.50	5.9	0.47	0.47	94
Control Dairy Kidney (1-22-B)	12.5	0.0	0.00	<0.20	<0.05	<0.05	
CGA-169374	12.5	0.62	0.05	0.55	0.044	0.044	89
CGA-169374	12.5	0.62	0.05	0.55	0.044	0.044	89
CGA-169374	12.5	1.25	0.10	1.20	0.096	0.096	96
CGA-169374	12.5	6.2	0.50	6.4	0.51	0.51	102
Control Chicken Lean Meat (1-23-B)	12.5	0.0	0.00	<0.20	<0.05	<0.05	
CGA-169374	12.5	0.62	0.05	0.48	0.038	0.038	76
CGA-169374	12.5	0.62	0.05	0.57	0.046	0.046	91
CGA-169374	12.5	1.25	0.10	1.13	0.091	0.091	91
CGA-169374	12.5	6.2	0.50	5.3	0.43	0.43	86

Comments: Recovery samples fortified prior to extraction.

*Dairy kidney and chicken lean meat ppm values are corrected for 80% moisture content by formula in Section II.J.2 of AG-544.

at/SEPTEMBER

RESIDUE RECOVERY REPORT

AG-A No. 11060-01

Page 6 of 7

Compound(s)		Substrate(s)					
CGA-169374		Control Chicken Eggs, Skin and Liver					
		Pesticide Added					
		CGA-169374					
SUBSTRATE	SAMPLE WEIGHT mg	AMOUNT ADDED		FOUND			
		ng	ppm	TOTAL		NET	
				ng	ppm*	ppm	%
Control Eggs (1-15-A)	12.5	0.0	0.00	<0.20	<0.05	<0.05	
CGA-169374	12.5	0.62	0.05	0.46	0.037	0.037	74
CGA-169374	12.5	0.62	0.05	0.51	0.041	0.041	81
CGA-169374	12.5	1.25	0.10	1.01	0.081	0.081	81
CGA-169374	12.5	6.2	0.50	5.2	0.41	0.41	83
Control Chicken Skin (1-20-A&1-24-A)	12.5	0.0	0.00	<0.20	<0.05	<0.05	
CGA-169374	12.5	0.62	0.05	0.58	0.046	0.046	92
CGA-169374	12.5	0.62	0.05	0.61	0.049	0.049	98
CGA-169374	12.5	1.25	0.10	1.19	0.095	0.095	95
CGA-169374	12.5	6.2	0.50	6.2	0.50	0.50	100
Control Chicken Liver (Commercial)	12.5	0.0	0.00	<0.20	<0.05	<0.05	
CGA-169374	12.5	0.62	0.05	0.61	0.049	0.049	97
CGA-169374	12.5	0.62	0.05	0.61	0.049	0.049	97
CGA-169374	12.5	1.25	0.10	1.15	0.092	0.092	92
CGA-169374	12.5	6.2	0.50	5.7	0.46	0.46	91

Comments: Recovery samples fortified prior to extraction.

*Chicken liver and eggs ppm values are corrected for 80% moisture content by formula in Section II.J.2 of AG-544.

at/SEPTEMBER

AG-A No. 11060-01

Page 7 of 7

Comments: Recovery samples fortified prior to extraction.

77/SEPTEMBER